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International Journal of Infectious Diseases 111 (2021) 5–9

Contents lists available at ScienceDirect

International Journal of Infectious Diseases

journal homepage: www.elsevier.com/locate/ijid

Low infectious risk of re-positive COVID-19 patients: a single-center study

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ARTICLE INFO

Article history:
Received 17 April 2021
Revised 27 July 2021
Accepted 9 August 2021

KEYWORDS:
SARS-CoV-2
COVID-19
Re-positive
Infectiousness

ABSTRACT

Objective: The aim of this study was to investigate the infectiousness of re-positive coronavirus disease 2019 (COVID-19) patients.
Methods: All nucleic acid testing (NAT) was performed using throat swabs, nasopharyngeal swabs, and anal swabs, which were tested by Fluorescent quantitative realtime PCR. Re-positive cases were defined as a discharged patient who re-tested positive by NAT. Micro-neutralization of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was performed based on the methods for severe acute respiratory syndrome (SARS) and Middle East respiratory syndrome (MERS) viruses. IgM and IgG against the N protein of SARS-CoV-2 were determined by ELISA.
Results: A total 255 (16.04%) of 1590 COVID-19 patients were re-positive. The re-positive cases were more likely to occur in patients in the 20–39 years age group and in patients with disease of moderate severity. Quantitative PCR showed that cycle threshold (Ct) values and viral loads were both far lower than in the hospitalized COVID-19 patients. The viral load in re-positive cases was very low. Viral culture of the samples from re-positive patients showed no cytopathic effect, and NAT of the culture medium of viral cultures all exhibited negative results.
Conclusion: The viral load in re-positive cases was very low; patients were not infectious and the risk of human-to-human transmission was extremely low. Discharged COVID-19 patients should undergo home health management for 3 weeks.
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1. Introduction

Coronavirus disease 2019 (COVID-19) is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) (Huang et al., 2020). Since it was first reported at the end of 2019, COVID-19 has become a global pandemic due to the high transmissibility of the virus (World Health Organization, 2020). According to the Diagnosis and Treatment Protocol for Novel Coronavirus Pneumonia (Trial Version 6) released by the National Health Commission of China (National Health Commission of China, 2020), all discharged COVID-19 patients should undergo a 14-day medical observation and quarantine. During this period, some discharged patients are found to re-test positive for SARS-CoV-2; these cases are defined as 're-positives'. Several studies have reported the occurrence of re-positives among discharged COVID-19 patients (Chen et al., 2020; Hoang et al., 2020; Jiang et al., 2020; Peng et al., 2020; Yuan J et al., 2020). It has been reported that about 7–10% of COVID-19 patients re-test positive for SARS-CoV-2 RNA by RT-PCR after discharge (Cao et al., 2020; Yuan B et al., 2020). However, the infectiousness of re-positive COVID-19 patients is largely unknown. Therefore, the aim of the current study was to investigate the infectiousness of re-positive COVID-19 patients.

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https://doi.org/10.1016/j.ijid.2021.08.019
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2. Methods

2.1. Study subjects

In accordance with the Diagnosis and Treatment Protocol for Novel Coronavirus Pneumonia (Trial Version 7) released by the National Health Commission of China (National Health Commission of China, 2020), a patient positive for both the *ORF* gene and *N* gene of SARS-CoV-2 by nucleic acid tests (NATs) was diagnosed with COVID-19. Re-positives were defined as a patient who re-tested positive by NAT after reaching the discharge standard (with negative results in two consecutive NATs). As of May 18, 2020, a total of 1590 COVID-19 patients had been diagnosed in the Guangdong Provincial Center for Disease Control and Prevention. Of these, 439 patients with neutralizing antibody (NAbs), IgM, and IgG data were included for serological analysis, including 237 re-positive cases and 202 non-re-positive cases.

2.2. Nucleic acid testing

The SARS-CoV-2 nucleic acid detection kit (Shanghai Berger Medical Technology Co., China) was used to detect the *ORF1ab* gene and *N* gene of SARS-CoV-2 using dual fluorescence reverse transcription PCR (RT -PCR), according to the manufacturer's protocol (Xu et al., 2020).

2.3. Virus isolation and culture

The throat swabs, nasal swabs, and stool specimens of re-positive cases were collected for virus isolation in a biosafety level 3 (BSL-3) laboratory using Vero-E6 cells. A 200-μl aliquot of the specimens from re-positive cases was added to a cell culture tube containing a single layer of Vero-E6 cells (80-95% confluency) using Dulbecco's Eagel medium (DMEM) (1% fetal calf serum (FCS), 100 U penicillin/ml, 100 μg streptomycin/ml, and 2 mM glutamine), followed by incubation for 3–7 days in a 37°C, 5% CO_2 incubator. When the cytopathic effect (CPE) was observed in all cells, the supernatant was aspirated into a new culture flask for subculture, and part of the supernatant was stored at −80°C.

2.4. Confirmation of negative COVID status before discharge

The negative COVID status was confirmed before discharge as follows: (1) body temperature returned to normal (<37.3 °C) for more than 3 days; (2) significant relief of respiratory symptoms; (3) computed tomography (CT) imaging showed that pulmonary inflammation was obviously absorbed; (4) two consecutive NATs showed negative results, and the interval between the two tests was at least 1 day. For the last sample tested before discharge, a throat swab, nasopharyngeal swab, and anal swab were tested by real-time fluorescence quantitative PCR; (5) after discharge, the patient needed to be home quarantined for 14 days and received a NAT on days 7 and 14 after discharge. Those with a negative result were released from home quarantine, and those with positive results were hospitalized again for treatment.

2.5. Serum sample preparation

For serum sample preparation, 3 ml of venous blood was collected in a vacuum blood collection tube, followed by incubation at room temperature for 20–120 min. After that, the tube was centrifuged at 1000 rpm for 20 min, and the resulting supernatant was collected as a serum sample.

2.6. Micro-neutralization of SARS-CoV-2

SARS-CoV-2 is a new virus, hence there is no established method of micro-neutralization test or threshold for a positive NAb titer. Diagnosis and treatment of Severe Acute Respiratory Syndrome Coronavirus (2003) established by China CDC. Since SARS-CoV-2 belongs to the betacoronaviruses, the micro-neutralization test method and threshold for positive NAb titer were established with reference to those of severe acute respiratory syndrome coronavirus (SARS-CoV) (Chinese Medical Association and Chinese Society of Traditional Chinese Medicine, 2003) and Middle East respiratory syndrome coronavirus (MERS-CoV) (GuangYu et al., 2013). All procedures of the micro-neutralization test were performed in a BSL-3 laboratory. First, 120 μl of diluted serum sample (1:4 to 1:1024 dilution) and 120 μl of SARS-CoV-2 virus stock (20SF014/vero-E6/3, titer 100× $TCID_{50}$ in 50µl) were added to the 96-well plate, followed by incubation in a 37°C, 5% CO_2 incubator for 2 h. An aliquot of 100 μl of the virus–serum mixture was added to a 96-well micro cell plate containing a Vero-E6 cell suspension (1×10^4 to 2×10^4 cells/0.1 ml), followed by incubation in a 37°C, 5% CO_2 incubator for 5–7 days. When the 100× $TCID_{50}$ in 50µl antigen control well showed complete CPE, the CPE results of each well were recorded. The reciprocal of the highest dilution of the serum that can protect 50% of the cells from CPE was defined as the titer of NAbs for SARS-CoV-2. A NAb titer ≥1:4 was defined as a positive result.

2.7. ELISA assay for IgM and IgG against the N protein of SARS-CoV-2

The recombinant nucleocapsid protein (N protein)-based ELISA kit (ZhongshanShengWu, Zhongshan, China) was used for the detection of IgM (N-IgM) and IgG (N-IgG) against the N protein of SARS-CoV-2 in four patients, according to the manufacturer's protocol. These four patients were originally close contacts of COVID-19 patients and then became COVID-19 patients after becoming infected. During quarantine management, it was found that the four patients had started to produce antibodies before a positive NAT result. Since the antibody can be detected at an early stage, the changes in antibody levels can be monitored during the entire course. Briefly, serum sample (100 μl, diluted 1:100) was added to the pre-coated plates, followed by incubation with horseradish peroxidase (HRP)-conjugated rN protein of SARS-CoV-2 (100 μl, 37°C for 30 min) and then 3,3′,5,5′-tetramethylbenzidine (TMB) substrate solution (50 μl). The reaction was terminated by adding 50 μl of 2 M sulfuric acid, and the absorbance value at 450 nm and 630 nm (A450–630) was determined. According to the kit instructions, the diagnostic cutoff value was 0.15 for IgM and 0.25 for IgG. When A450–630 was greater than or equal to the cutoff value, the test was considered positive. Three replicates of each sample were included on each plate.

2.8. Statistical analysis

Continuous data were recorded as the mean ± standard deviation (SD). For comparisons between the two groups, the Student independent *t*-test was used. The Mann–Whitney *U*-test was used if the normality of continuous variables was not assumed. Categorical variables were recorded as the number and percentage and were compared using the Chi-square test or Fisher's exact test (if an expected value ≤5 was found). For comparisons between two dependent dichotomous variables, the McNemar test was used. Logistic regression models were used to investigate the association between independent variables and being re-positive. A two-tailed *P*-value <0.05 was recognized as significant different for each test. All analyses were performed using IBM SPSS Statistics version 25 (IBM Corp. Armonk, NY, USA).

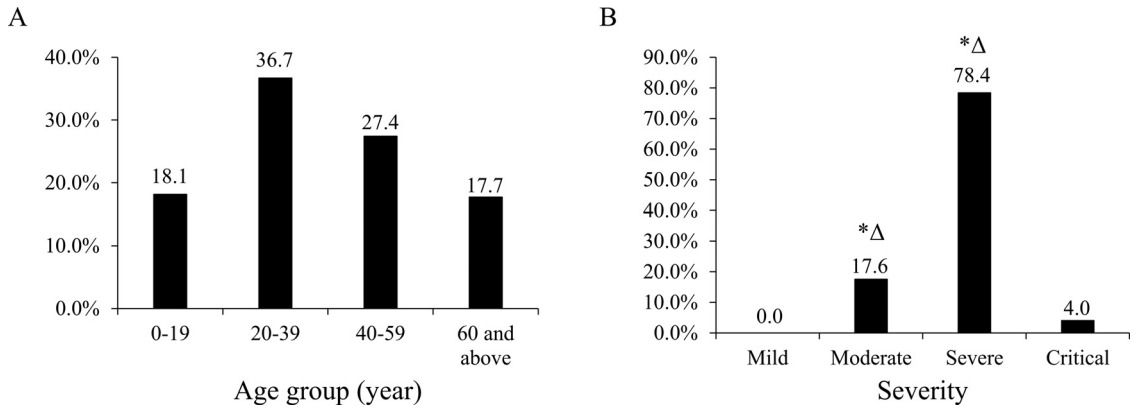

Figure 1. The re-positive rate in the regional population by (A) age group, and (B) severity group. $^{*}P < 0.05$ compared to the mild group; $^{\Delta}P < 0.05$ compared to the critical group.

Table 1
Demographic and clinical characteristics of the patients

Parameters	Non-re-positive (n = 202)	Re-positive (n = 237)	All (n = 439)	P-value
Sex				0.105
Male	102 (50.50%)	138 (58.23%)	240 (54.67%)	
Female	100 (49.50%)	99 (41.77%)	199 (45.33%)	
Age (years)	41.86 ± 19.84	38.63 ± 19.57	40.09 ± 19.74	0.090
Age group (years)[a]				0.392
1–19	29 (14.87%)	43 (18.14%)	72 (16.67%)	
20–39	63 (32.31%)	87 (36.71%)	150 (34.72%)	
40–59	67 (34.36%)	65 (27.43%)	132 (30.56%)	
≥60	36 (18.46%)	42 (17.72%)	78 (18.06%)	
Severity[a]				<0.001
No symptoms	27 (16.46%)	0 (0.00%)	27 (7.44%)	
Mild	20 (12.20%)	35 (17.59%)	55 (15.15%)	
Moderate	90 (54.88%)	156 (78.39%)	246 (67.77%)	
Severe and critical	27 (16.46%)	8 (4.02%)	35 (9.64%)	
Time after symptom onset (days)	41.66 ± 20.07	55.30 ± 16.57	49.02 ± 19.47	<0.001
Quarantine days	4.29 ± 4.76	4.44 ± 4.63	4.38 ± 4.66	0.863

Data presented as the number and percentage, or as the mean ± standard deviation.
[a] Age and disease severity data were missing for some patients, hence the sum for these two variables is not equal to the sample size of each group.

3. Results

3.1. Epidemiology and analysis of viral load in re-positive COVID-19 patients

As of May 18, 2020, a total of 1590 COVID-19 cases had been diagnosed in the study center; of these, 255 (16.04%) were re-positive cases. The male to female sex ratio was 1:1 and the median age of the patients was 34 years (range 0.25–86 years). The re-positive rate decreased significantly with increasing age (Figure 1A) and increasing severity of the disease (except for critical patients, Figure 1B) (all $P < 0.05$).

A NAT was performed for 223 re-positive patients. Of these, 64.13% (143/223, cumulative) were re-positive within 7 days after discharge and 99.55% (222/223, cumulative) were re-positive within 14 days after discharge; the range was from 1 to 64 days (median 10 days). Of the re-positive cases, 62.78% (140/223, cumulative) turned negative within 2 weeks after becoming re-positive and 71.30% (159/223, cumulative) turned negative within 4 weeks after becoming re-positive. The time range of turning negative was from 6 to 75 days (median 20 days).

Quantitative fluorescence PCR revealed a mean Ct value for the virus *ORF1ab* gene in 87 re-positive patients of 37 (range 32 to 40). The mean viral load was 1138 copies/ml (range 18 to 6430 copies/ml), which was far lower than those of the hospitalized COVID-19 patients (critical, 3.0×10^7 copies/ml; severe, 1.1×10^7 copies/ml; moderate, 7.4×10^6 copies/ml; mild, 4.9×10^6 copies/ml; no symptoms, 6.0×10^5 copies/ml).

3.2. Serological analysis

A total of 439 patients (mean age 40.09 ± 19.74 years, 240 male and 199 female) with NAbs, IgM, and IgG data were included for serological analysis, including 237 re-positive cases and 202 non-re-positive cases. As indicated in Table 1, the re-positive group had significantly less severe and critical disease cases and a longer sampling time after symptom onset as compared with the non-re-positive group (both $P < 0.05$).

As shown in Table 2, the level and positive rate of IgM were significantly lower in the re-positive group than in the non-re-positive group (both $P < 0.05$). No significant difference in NAbs or IgG results was found between the groups.

Micro-neutralization showed positive results in 222 (93.67%) re-positive patients and 184 (91.09%) non-re-positive patients ($P = 0.307$). For the re-positive group, the antibody could be tested positive at a median of 58 days (range 1 to 85 days) after onset, and the mean titer was 1:34.4 (range negative to 1:2048). For the non-re-positive group, the antibody could be tested positive at a median of 42 days (range 3 to 85 days) after onset, and the mean titer was 1:109.9 (range negative to 1:1024). The titer did not differ significantly between the re-positive and non-re-positive groups ($P = 0.477$).

Table 2
Serological analysis of re-positive and non-re-positive patients

Parameters	Non-re-positive (n = 202)	Re-positive (n = 237)	All (n = 439)	P-value
NAbs				
Negative	18 (8.91%)	15 (6.33%)	33 (7.52%)	0.307
Positive	184 (91.09%)	222 (93.67%)	406 (92.48%)	
IgM level	0.33 ± 0.67	0.19 ± 0.56	0.26 ± 0.62	0.017
Negative	141 (69.80%)	190 (80.17%)	331 (75.40%)	0.012
Positive	61 (30.20%)	47 (19.83%)	108 (24.60%)	
IgG level	1.17 ± 0.93	1.03 ± 0.79	1.10 ± 0.86	0.099
Negative	27 (13.37%)	44 (18.57%)	71 (16.17%)	0.140
Positive	175 (86.63%)	193 (81.43%)	368 (83.83%)	

NAbs, neutralizing antibodies.

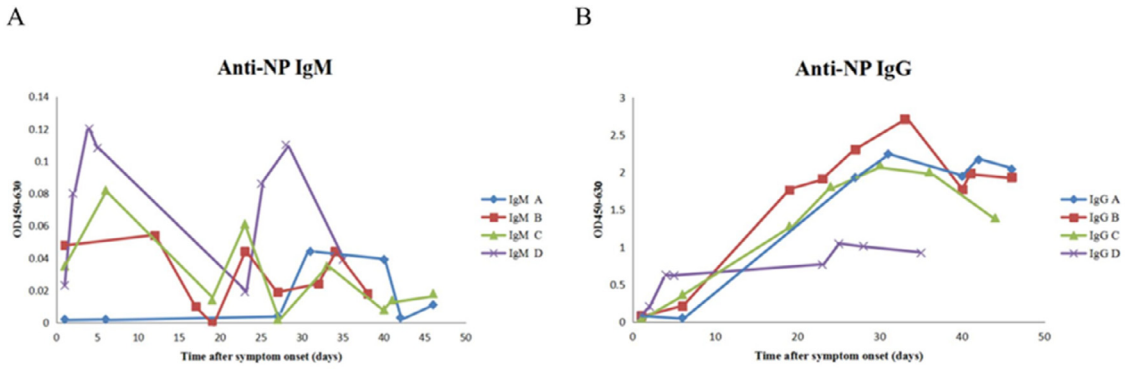

Figure 2. The levels of (A) anti-NP-IgM, and (B) anti-NP IgG in four patients (patients A, B, C, and D) over time. (NP, anti-nucleocapsid protein.)

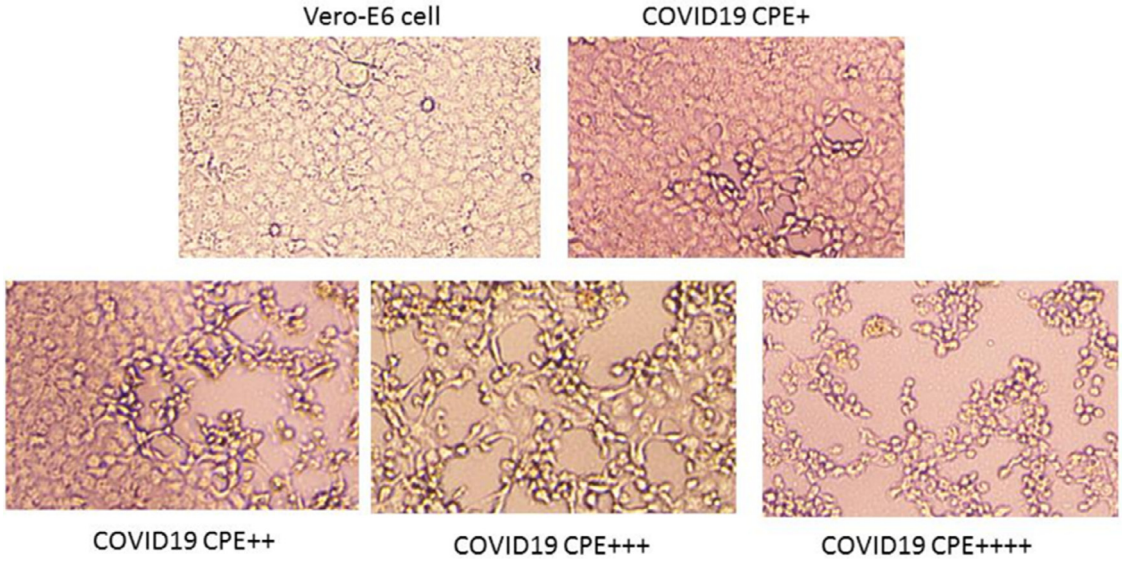

Figure 3. The cytopathic effect (CPE) levels in Vero-E6 cells after being infected with virus in the neutralization study: + 25%, ++ 50%, +++ 75%, ++++ 100%.

Continuous sampling on $OD_{450–630}$ for N-IgM and N-IgG protein was performed for four re-positive cases. As shown in Figure 2, no obvious trend was found in N-IgM. Patient D was found to be N-IgG positive on the fourth day after onset, while the other three patients were positive on the sixth day after onset. The $OD_{450–630}$ for N-IgG of patient D increased intensely on days 1, 2, and 4 and then remained stable. The $OD_{450–630}$ for N-IgG of the other three patients reached a high peak on day 30 to 35, decreased on day 35 to 40, and plateaued after day 45.

3.3. Viral load in re-positive patients

The gene copy of ORF was 1.6×10^4 copies/ml (range 6.3×10^5 to 17×10^5 copies/ml) and of N was 1.6×10^4 copies/ml (range 6.3×10^5 to 17×10^5 copies/ml). These results were far lower than the results before turning re-positive.

No cytopathic effect was found and no virus was isolated with nasopharynx and stool samples of 92 re-positive patients infection Vero-E6 cell. SARS-COV2 gene was not detected on qPCR of the culture medium of vero-E6 cell incubated with patients specimen. This might have been due to the low viral loads in the samples (Figure 3).

4. Discussion

The term ‘re-positive’ is defined as the detection of SARS-CoV-2 nucleic acid in the body again after a COVID-19 patient has re-

covered and been discharged. Re-positive cases have been reported in several countries, including China (Wong et al., 2021), Japan (Schneider et al., 2020), and Brunei (Wong et al., 2020). The results of the current study showed that re-positive cases were more likely to occur in patients in the age group of 20–39 years and in patients with disease of moderate severity. Continuous monitoring and sampling of re-positive patients revealed that 64.13% of cases re-tested positive by NAT within 7 days after discharge and 99.55% of cases were re-positive within 14 days. Of all the re-positive cases, 71.30% turned negative within 4 weeks after becoming re-positive. Therefore, for discharged patients, 14 days of medical isolation and monitoring are required, and re-positive cases need to be monitored continuously for at least 14 days until turning negative.

In this study, fluorescence qPCR detection of the *ORF1ab* gene showed that the mean Ct value and the mean viral load were both far lower in the re-positive cases than in the hospitalized COVID-19 cases. In the re-positive cases, no CPE was observed during cell passage for three generations. Furthermore, NATs all exhibited negative results for the culture medium of viral culture. These results suggest that the viral load of the re-positive cases was very low, they were not infectious and the risk of human-to-human transmission was extremely low. Consistent with these findings, a Japanese study by Ogawa et al. (Schneider et al., 2020) analyzed 15 healthcare personnel who had contact exposure and aerosol exposure to re-positive COVID-19 patients, and found that all healthcare personnel were negative for SARS-CoV-2 in both the serological analysis (IgG) and PCR tests (nasopharyngeal swabs) on day 10. In addition, none of them had any symptoms or apparent infection during 10 days of active isolation. Likewise, a study by Wong et al. in Brunei reported that all 111 close contacts exposed to the 21 re-positive cases had a negative RT-PCR result for SARS-CoV-2 (Wong et al., 2020). All of these results suggest a low infectivity potential in re-positive COVID-19 patients.

There are some limitations to this study. First, the sample size was relatively small. In addition, due to the retrospective nature of this study, not all patients had complete data. In the future, a large prospective trial should be conducted to validate the findings of this study.

In summary, this study suggests that although COVID-19 patients recovered and the viral load was too low to be isolated, SARS-CoV-2 may still be excreted at a low level. Therefore, continuous monitoring of the viral load and antibody levels should be simultaneously considered in order to develop epidemic prevention policies. Serological analysis is especially important for patients with an extremely low viral load. We recommend that discharged COVID-19 patients should undergo home health management for 3 weeks. If no symptoms are presented during the 3 weeks, the patients can return to normal life after clinical consultation.

Declarations

Funding source: This study was funded by the NIAID Center of Excellence for Influenza Research and Surveillance (HHSN2722014000006C), the Key Research and Development Program of Guangdong Province (2019B111103001), and the Natural Science Foundation of Guangdong Province (2019A1515012121).

Ethical approval: Ethical approval and consent to participate were not required due to the retrospective nature of this study.

Conflict of interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

Cao H, Ruan L, Liu J, Liao W. The clinical characteristic of eight patients of COVID-19 with positive RT-PCR test after discharge. J Med Virol 2020;92:2159–64. doi:10.1002/jmv.26017.

Chen Y, Bai W, Liu B, Huang J, Laurent I, Chen F, et al. Re-evaluation of retested nucleic acid-positive cases in recovered COVID-19 patients: Report from a designated transfer hospital in Chongqing, China. J Infect Public Health 2020;13:932–4. doi:10.1016/j.jiph.2020.06.008.

Chinese Medical Association;, Chinese Society of Traditional Chinese Medicine. [Infectious atypical pneumonia (SARS) diagnosis and treatment plan]. Natl Med J China 2003;83:1731–52.

GuangYu Z, LanYing D, CuiQing M, Ye L, Lin L, Poon VK, et al. A safe and convenient pseudovirus-based inhibition assay to detect neutralizing antibodies and screen for viral entry inhibitors against the novel human coronavirus MERS-CoV. Virol J 2013;10:266.

Hoang VT, Dao TL, Gautret P. Recurrence of positive SARS-CoV-2 in patients recovered from COVID-19. J Med Virol 2020 jmv.26056. doi:10.1002/jmv.26056.

Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020;395:497–506. doi:10.1016/S0140-6736(20)30183-5.

Jiang M, Li Y, Han M, Wang Z, Zhang Y, Du X. Recurrent PCR positivity after hospital discharge of people with coronavirus disease 2019 (COVID-19). J Infect 2020;81:147–78. doi:10.1016/j.jinf.2020.03.024.

National Health Commission of China. Diagnosis and Treatment Protocol for Novel Coronavirus neumonia (Trial Version 6) 2020.

Peng J, Wang M, Zhang G, Lu E. Seven discharged patients turning positive again for SARS-CoV-2 on quantitative RT-PCR. Am J Infect Control 2020;48:725–6. doi:10.1016/j.ajic.2020.03.017.

Schneider KN, Correa-Martínez CL, Gosheger G, Rickert C, Schorn D, Mellmann A, et al. Assessing the spreading potential of an undetected case of COVID-19 in orthopaedic surgery. Arch Orthop Trauma Surg 2020. doi:10.1007/s00402-020-03516-1.

Wong CL, Lei SK, Lei CI, Lo IL, Lam C, Leong IH. Re-positive of SARS-CoV-2 test is common in COVID-19 patients after hospital discharge. Data from high standard post-discharge quarantined patients in Macao SAR, China. PeerJ 2021;9. doi:10.7717/peerj.11170.

Wong J, Koh WC, Momin RN, Alikhan MF, Fadillah N, Naing L. Probable causes and risk factors for positive SARS-CoV-2 test in recovered patients: Evidence from Brunei Darussalam. J Med Virol 2020;92:2847–51. doi:10.1002/jmv.26199.

World Health Organization. Coronavirus disease (COVID-19) Global epidemiological situation. 2020.

Xu XW, Wu XX, Jiang XG, Xu KJ, Ying LJ, Ma CL, et al. Clinical findings in a group of patients infected with the 2019 novel coronavirus (SARS-Cov-2) outside of Wuhan, China: Retrospective case series. BMJ 2020;368. doi:10.1136/bmj.m606.

Yuan B, Liu HQ, Yang ZR, Chen YX, Liu ZY, Zhang K, et al. Recurrence of positive SARS-CoV-2 viral RNA in recovered COVID-19 patients during medical isolation observation. Sci Rep 2020;10. doi:10.1038/s41598-020-68782-w.

Yuan J, Kou S, Liang Y, Zeng J, Pan Y, Liu L. PCR Assays Turned Positive in 25 Discharged COVID-19 Patients. Clin Infect Dis 2020. doi:10.1093/cid/ciaa398.